

VICAM
AFLA-V QUANTITATIVE AFLATOXIN TEST KIT

Table of Contents	Page
GENERAL INFORMATION	1
PREPARATION OF TESTING MATERIALS.....	2
SAMPLE PREPARATION AND EXTRACTION PROCEDURES	3
TEST PROCEDURES	3
SUPPLEMENTAL ANALYSIS	4
REPORTING AND CERTIFYING TEST RESULTS	4
STORAGE CONDITIONS AND PRECAUTIONS.....	4
EQUIPMENT AND SUPPLIES	4
REVISION HISTORY	5

GENERAL INFORMATION

Afla-V is VICAM's rapid lateral flow strip test for the quantitation of total aflatoxins in the range of 5-100 ppb.

The instructions presented in this document cover only the procedure for performing the analytical test for official inspections. For questions regarding this procedure, contact Dr. Ajit Ghosh of the Technology and Science Division by phone at 816-891-0417 or email at Ajit.K.Ghosh@usda.gov.

Refer to the current policies and/or instructions issued by the Policies, Procedures, and Market Analysis Branch (PPMAB) of the Field Management Division for information on use of this test kit in official inspections including sampling, general sample preparation (e.g., grinding and dividing), reporting and certification of test results, laboratory safety, and hazardous waste management. For questions regarding these policies and/or instructions, contact Patrick McCluskey of PPMAB by phone at 816-659-8403 or email at Patrick.J.McCluskey@usda.gov.

Approved Test Kit Information

Test Kit Vendor:	<i>Vicam, 800-338-4381 or 508-482-4935</i>
Test Kit Name:	Afla-V
Product Number:	176002071
Effective Date of Instructions:	01/07/2015
Instructions Revision Number	1
Conformance Range:	5 – 100 ppb
Number of Analyses to Cover Conformance Range:	2
Type of Service:	Quantitative
Supplemental Analysis:	No
Approved Commodities:	Corn
Extraction method:	Blend 50 gram sample with 250 milliliters (mL) of 70% MeOH/30% deionized water (v/v) for 1 minute.
Test Format:	Lateral flow strip
Detection Method:	Vertu Reader, Model Vertu

PREPARATION OF TESTING MATERIALS

Purchasing of Extraction Solution:

- (1) The extraction solvent used in the Afla-V quantitative test kit method is a methanol/water mixture consisting of 70 percent methanol (reagent grade or better) and 30 percent water (distilled or deionized). The solution can be purchased from VICAM, part number 100000217.

Preparation of Extraction Solution:

- (1) Alternatively a 70 percent methanol/30 percent deionized or distilled water solution (v/v) can be made as follows.
- (2) Using a graduated cylinder, measure 700 mL of methanol (ACS reagent grade or better) and place it into a glass bottle or carboy.
- (2) Add 300 mL deionized or distilled water to the methanol and shake vigorously until completely mixed.
- (3) Label the container stating the mixture (70 percent methanol: 30 percent water), date of preparation and initials of technician that prepared the solution.
- (4) Store this solution at room temperature in a tightly closed container.
- (5) To prepare smaller or larger amounts of solution use the ratio of 7 parts methanol to 3 parts of deionized or distilled water.

Preparation of Reader:

- (1) Turn on the power for Vertu-incubator. Pre-set temperature at 30°C and let warm up until 30°C is displayed on the incubator.
- (2) Turn on the Vertu reader. Using the arrow keys, move the cursor to “measure” on the display, and then press the center key. You will then hear 3 beeps.
- (3) Calibrate the Vertu reader daily by scanning in the bar code for the lot of strips being used. You will hear a beep when the bar code is accepted. Make sure the lot number displayed on the reader is the same lot number as the strips and is for the incubation method at 30°C.

SAMPLE PREPARATION AND EXTRACTION PROCEDURES

Standard Extraction Procedure for Corn

- (1) Weigh 50 gram (± 0.2 g) ground sample into a blender jar.
- (2) Add to the blender jar 250 mL of 70% MeOH.
- (3) Cover the blender jar and blend at high speed for 1 minute.
- (4) Filter the extract through VICAM filter paper (order # 600001106) into a clean extraction tube. Cap the tube as MeOH may evaporate if it stays open long than 2 minutes. This filtered-extract can be used for next 2 hours if the cap of the tube is kept closed.

TEST PROCEDURES

a. Test Procedure for 5 to 40 ppb Quantitation Range:

- (1) Add 100 μ L of the AFLA-V diluent into the strip test vial.
- (2) Add 100 μ L of the filtered extract to the vial. Cover and mix well by vortexing.
- (3) Place above mixture from step 2 and an AFLA-V strip cassette (circular opening side in first), into the Vertu-incubator. Close the incubator plastic cover and incubate at 30°C for 4 minutes.
- (4) Transfer 100 μ L of this pre-warmed solution from step 3 to the circular opening on the pre-warmed AFLA-V strip at a rate of about 1 drop per second. Be sure to hold the pipet vertically. The pipet tip can touch the bottom of cassette.
- (5) Close the incubator plastic cover and allow the strip to develop at 30°C for 4 minutes.
- (6) Insert the AFLA-V strip into the Vertu reader (circular opening side in first).
- (7) Press the center key on the reader to read the strip.
- (8) To print the result, move the cursor to "P" in the upper left hand corner of the display using the arrow keys then press the center key.
- (9) To run the next sample, move the cursor to "NT" using the arrow keys and press the center key. If you are using the same lot of strips, you are now ready to insert the next sample. If you are using a different lot of strips, you will need to scan in the new bar code.

b. Test Procedure for 40 to 100 ppb Quantitation Range:

- (1) If the reader displays "> Range" (40 ppb or more), dilute filtered sample extract from step 4 (of

Standard Extraction Procedure for Corn) one to five with 70% MeOH (100 µL extract + 400 µL 70% MeOH).

- (2) With the diluted extract repeat steps **1** to **9** (of Test Procedure for 5 to 40 ppb Quantitation Range with the diluted extract).
- (3) Multiply the displayed result by **4** to obtain the true level of contamination.

SUPPLEMENTAL ANALYSIS

This test kit does not provide any supplemental analysis

REPORTING AND CERTIFYING TEST RESULTS

Refer to the current instructions issued by the Policies, Procedures, and Market Analysis Branch of the Field Management Division for reporting and certification of test results. For questions regarding these instructions, contact Patrick McCluskey (816-659-8403 or Patrick.J.McCluskey@udsa.gov).

STORAGE CONDITIONS AND PRECAUTIONS

a. Storage Conditions:

- (1) Test Strip must be stored in refrigerator at 4° - 8° C (39° - 46° F). Strips must be brought to room temperature before using (may take about 45 minutes).

b. Precautions.

- (1) Do not use the test kits beyond the expiration date.
- (2) Prolonged exposure of test strips to high temperatures may adversely affect the test results.
- (3) Do not open the test strip package until ready to use the strips.

EQUIPMENT AND SUPPLIES

a. Supplied with the Test Kit.

- (1) Afla-V Kit (25 tests) part #176002071
- (2) AFLA-V KIT CONTENTS
- (3) Afla-V Strip Tests

- (4) Afla-V Diluent, 6 mL
- (5) Micro-Pipette Tips, 100 µL
- (6) Strip Test Vials, 1.5 mL
- (7) Filter Paper
- (8) Extraction Tubes, 40 mL
- (9) Afla-V Barcode Set
- (10) Afla-V Instruction Guide

b. Equipment and Materials Needed but not Supplied with the Test Kit.

- (1) Vertu Reader, 110V & 220V with barcode scanner, part #725000574
- (2) Custom V-incubator w/block, 110V & 220V, part#600001330
- (3) Blender w/ Stainless Steel 1 Qt. Blender Jar 110V, part #20200
- (4) Vertu Printer, 110V & 220V, part #725000577
- (5) Digital Timer, part #G4036
- (6) Micro-Pipettor, 100 µL, part #600001108
- (7) Filter Funnel, 65 mm, (10 packs), part#36020
- (8) Strip Test Vial Rack, part #600001107
- (9) Graduated Cylinder, 250 mL, part #20250
- (10) 70% Methanol/30% Water (v/v) - 4L Cubitainer, part#100000217
- (11) Alternatively, 70% Methanol/30% Water (v/v) can be made from ACS grade or better methanol and deionized or distilled water as described in the section “**Preparation of Extraction Solution.**”

REVISION HISTORY

Revision 1 (01/07/2015)

- Correct Acronym of Policies, Procedures, and Market Analysis Branch (PPMAB) has been used.
- Phone number of Patrick McCluskey (816-659-8403) has been corrected.

Revision 0 (12/02/2014)